Distance Dependent Interhelical Couplings of Organic Rods Incorporated in DNA 4-Helix Bundles

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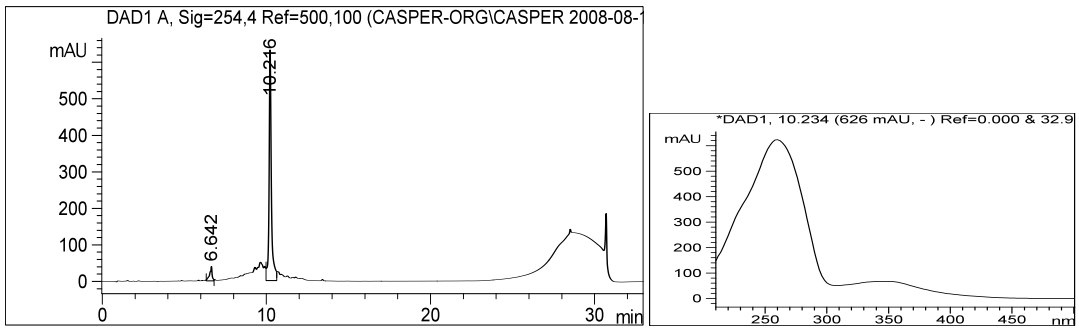
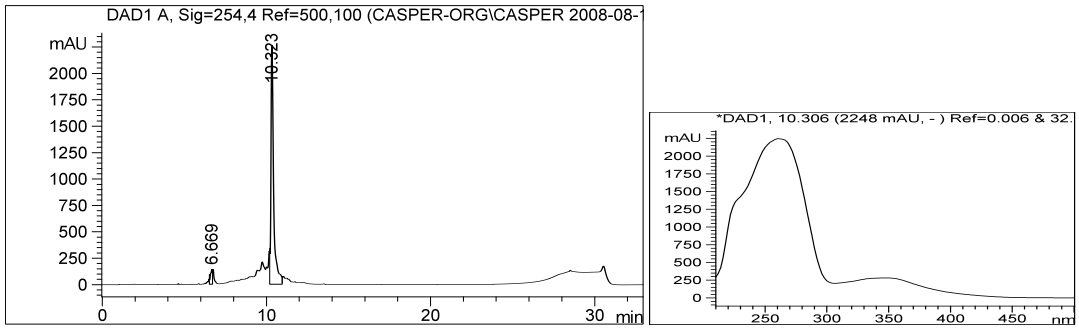
Figure S1. HPLC chromatograms and UV-VIS spectra of DNA conjugates.

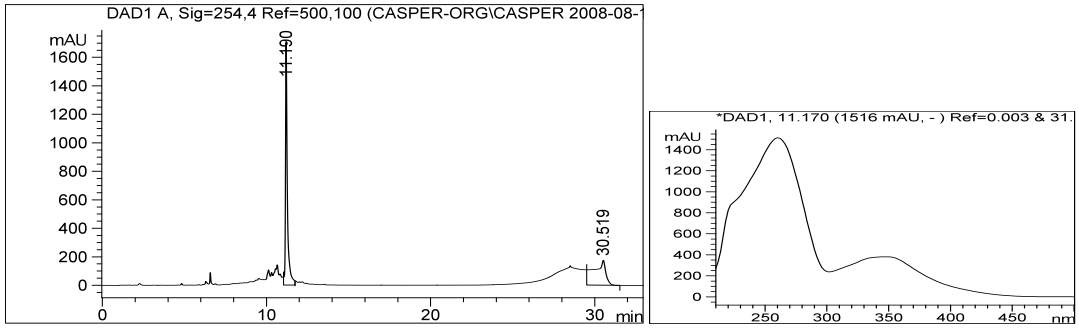
A) Conjugate **O2**



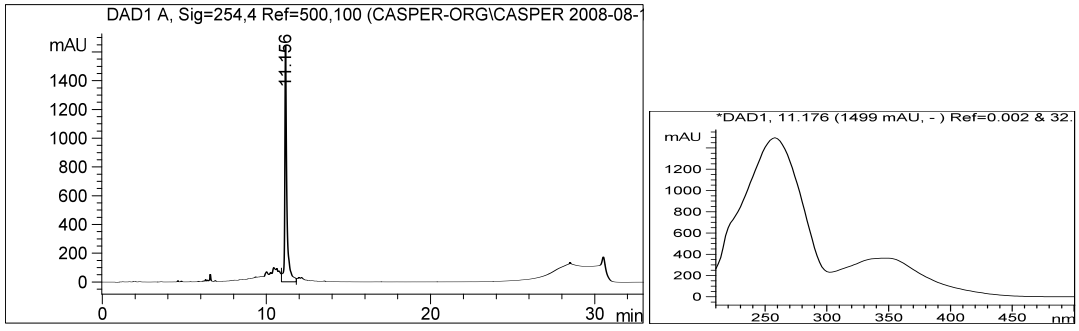
B) Conjugate **O4**



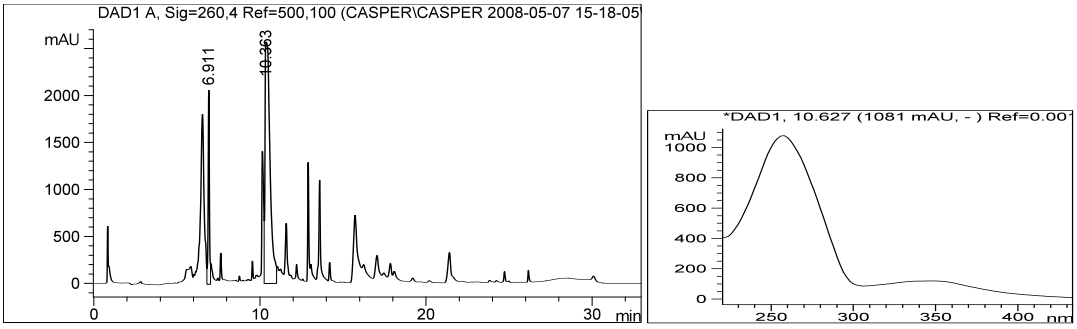
C) Conjugate **O7b**



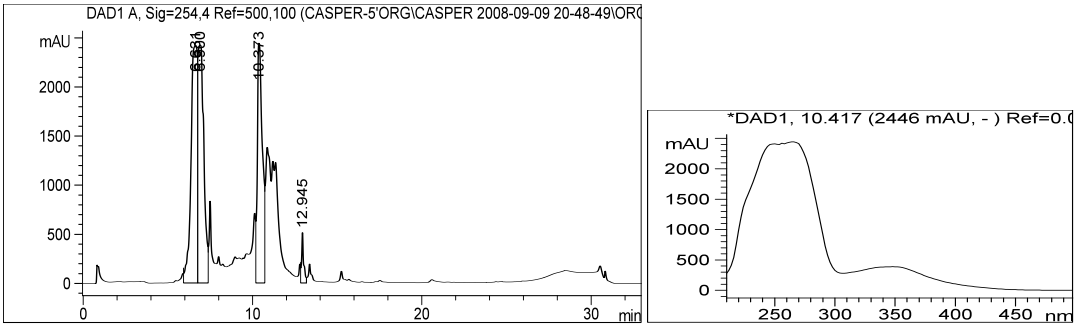
D) Conjugate **O8b**



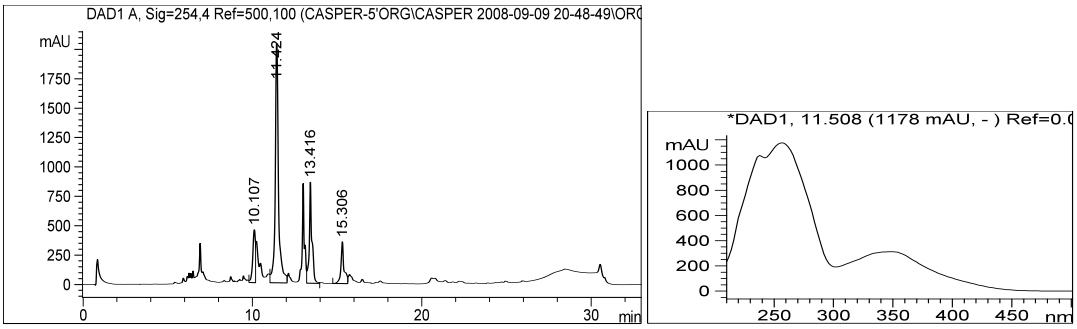
E) Conjugate **O1**



F) Conjugate **O3**



G) Conjugate **O7a**



H) Conjugate **O8a**

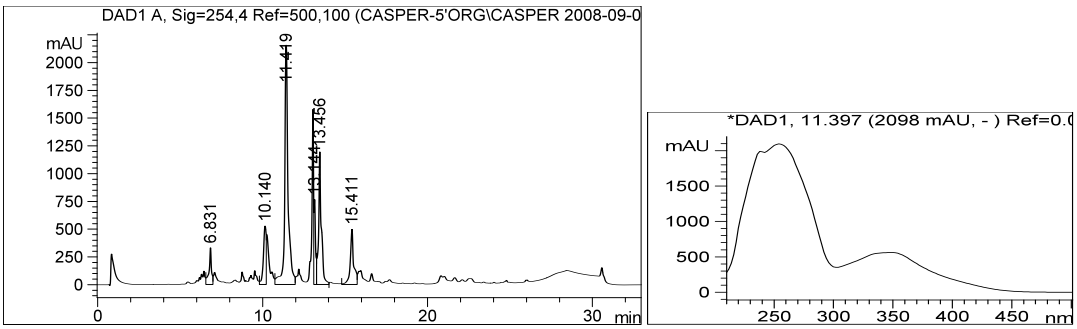
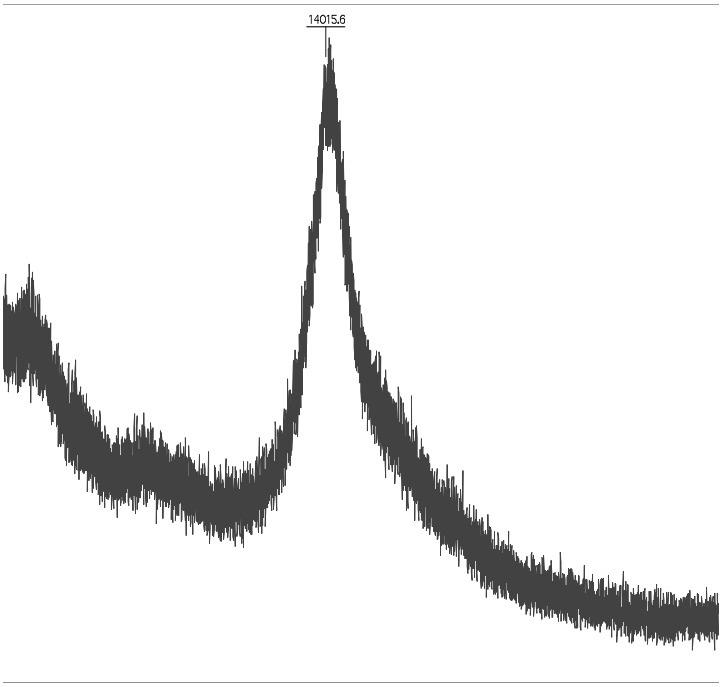
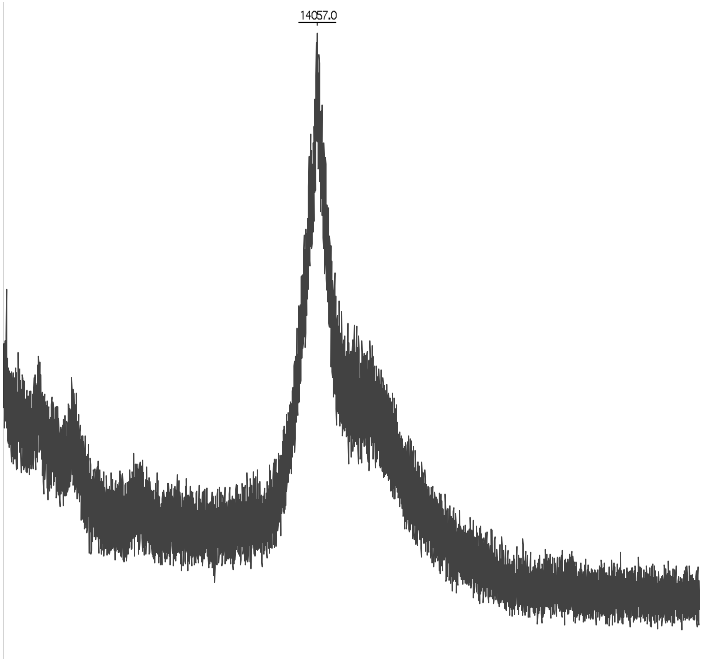


Figure S2. MALDI-TOF MS of the HPLC purified modified ODNs.

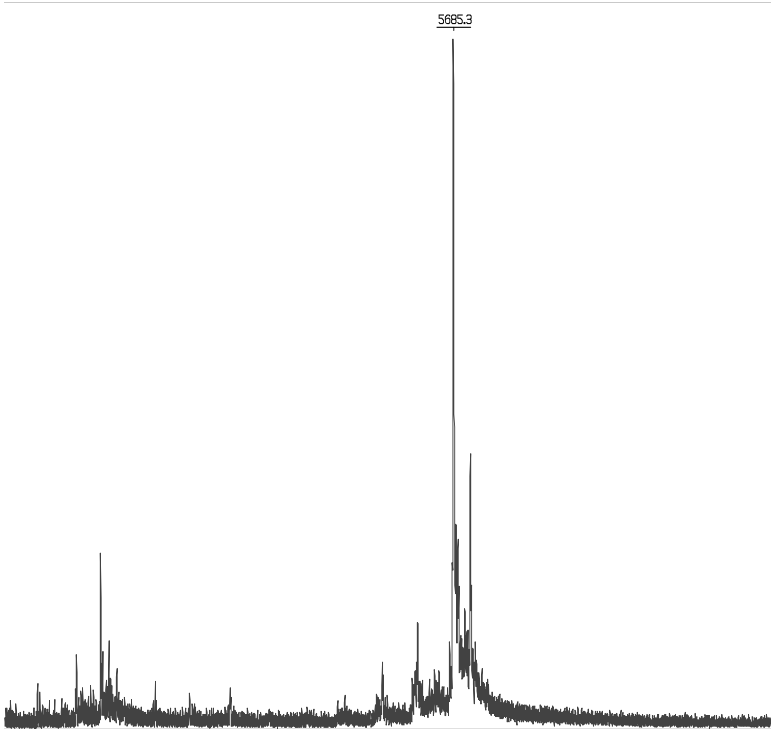
A) Conjugate **O2**: Calcd 14010 Found 14015



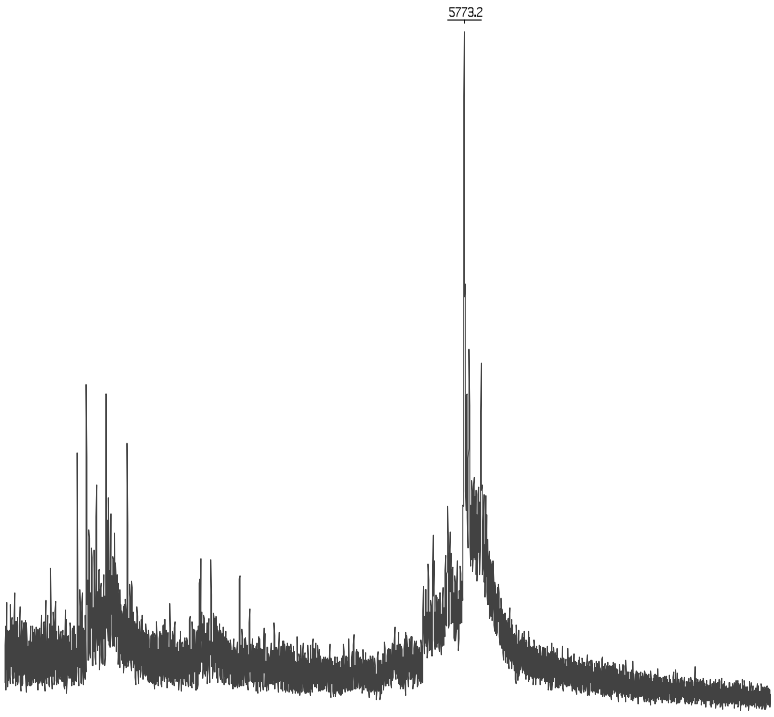
B) Conjugate **O4**: Calcd 14070 Found 14057



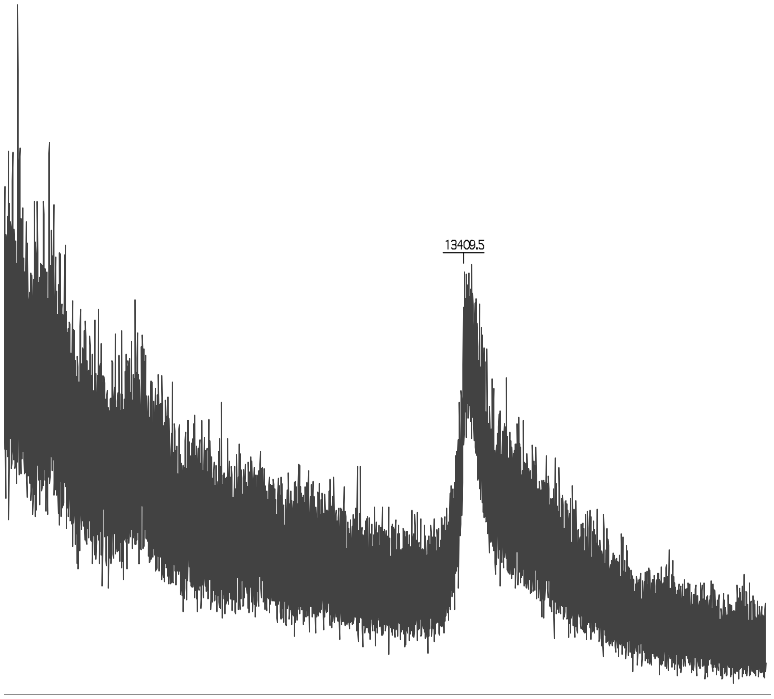
C) Conjugate **O7b**: Calcd 5986 Found 5985. The peak at [M+138] corresponds to the product + one matrix molecule.



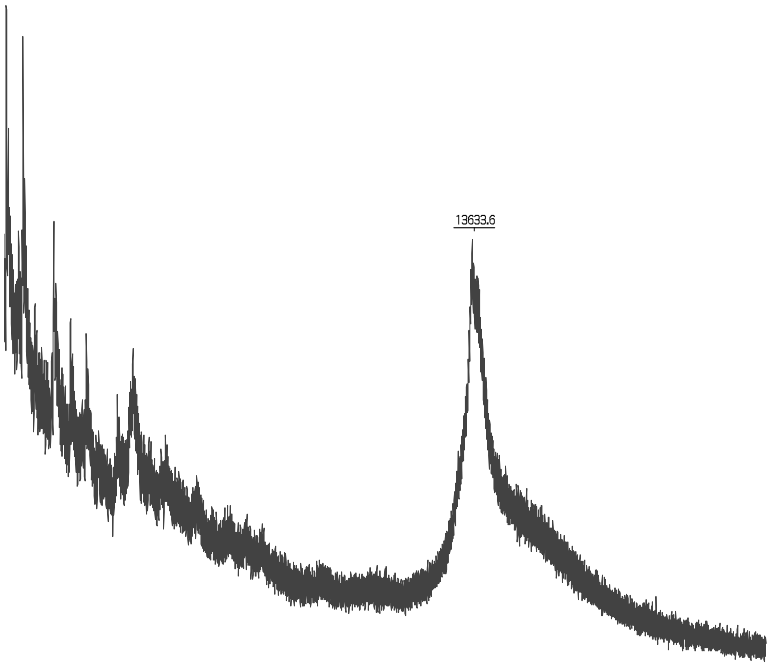
D) Conjugate **O8b**: Calcd 5782 Found 5773. The peak at [M+138] corresponds to the product + one matrix molecule.



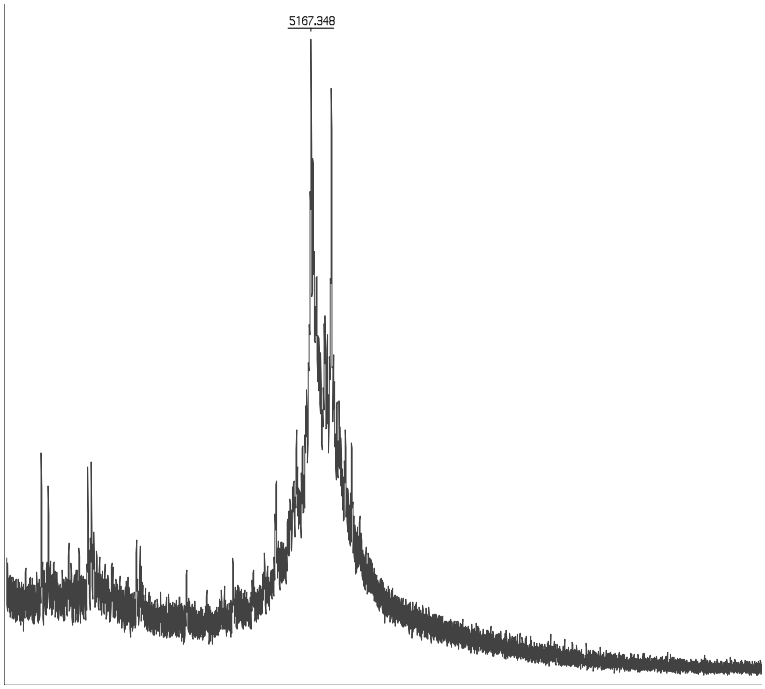
E) Conjugate **O1**: Calcd 13414 Found 13410.



F) Conjugate **O3**: Calcd 13634 Found 13634



G) Conjugate **O7a**: Calcd 5162 Found 5167. The peak at [M+138] corresponds to the product + one matrix molecule.



H) Conjugate **O8a**: Calcd 5258 Found 5258. The peak at [M+138] corresponds to the product + one matrix molecule.

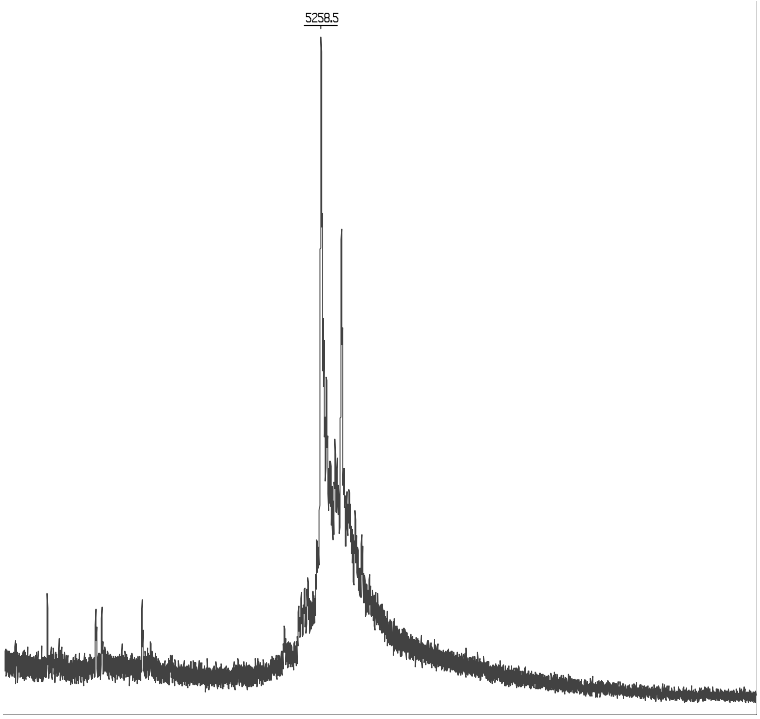
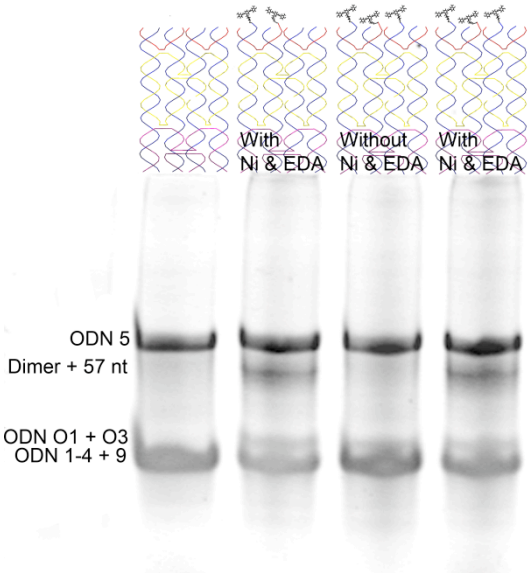
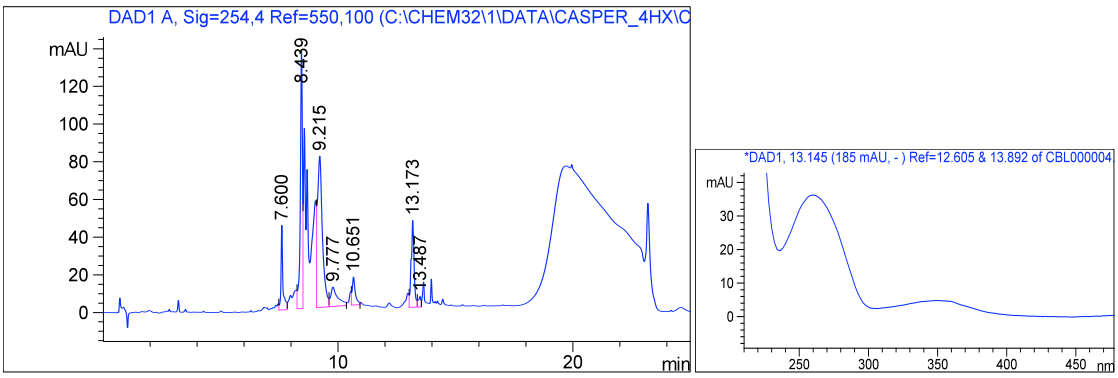


Figure S3. 4-HB templated nickel-salen couplings. (A) 9% denaturative PAGE analysis of 4-HBs, accumulated to one fifth of the original volume on filter cups before gel loading. (B) RP-HPLC chromatogram of the 4-HB with **O1** and **O7a** treated with nickel(II)chloride and EDA. (C) MALDI-TOF analysis of the partly purified dimer. (D) PAGE analysis of the partly purified dimer.

A) Lane 1 (unmodified 4-HB). Lane 2 (4-HB with **ODN O1** and **O7a** without coupling reagents). Lane 3 (4-HB with **ODN O1** and **O7a** without coupling reagents). Lane 4 (4-HB with **ODNO1** and **O7a** with coupling reagents).

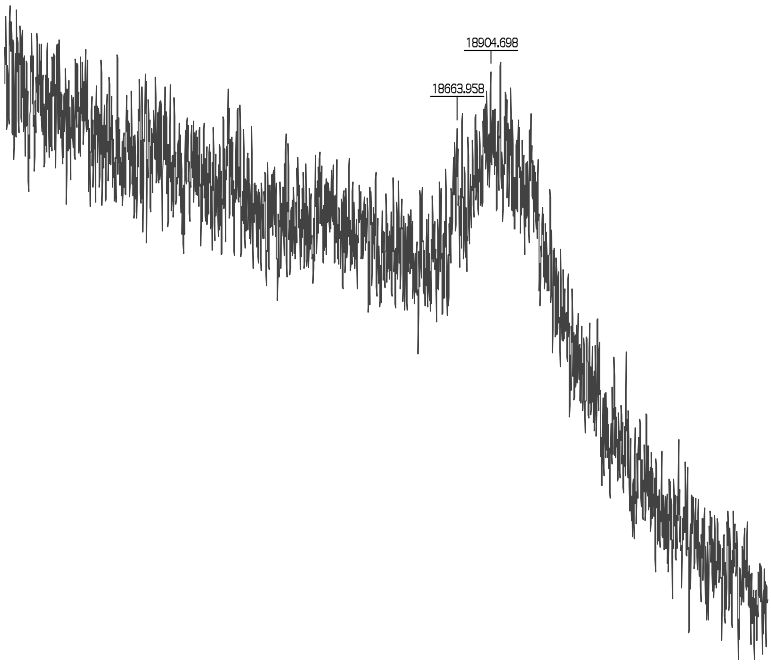


B) RP-HPLC chromatogram of a 4-HB with **ODN O1** and **ODN O7a** treated with nickel(II)chloride and EDA for 4 hours. The initial eluent is pure 0.1 M TEAA and is changed stepwise to separate the unmodified DNA strands (elutes from 7 to 10 minutes) from the organic modified DNA strands (eluted at 13 minutes). The peak at 13 minutes contains a mixture of monomers and the dimer. The UV-VIS spectrum of the collected sample at 13 minutes is shown. The purpose with this stepwise procedure is to remove all unmodified DNA strands and thereby enable the identification of the dimer. The fraction at 13 minutes is analyzed with MALDI-TOF MS and PAGE analysis (see Figure S3C and D)



The stepwise method: The acetonitrile percentage is increased from 0 to 20% in the first 10 minutes. From 10 to 17 minutes is the acetonitrile percentage held constant at 70%.

C) MALDI-TOF analysis of the HPLC fraction collected at 13 minutes, according to the HPLC spectrum in Figure S3B. The peak is very broad and it is therefore difficult to find an exact mass. Expected mass of the **O1**-**O7a** dimer: 18573. Found: 18633.



D) PAGE analysis. An 4-HB with **ODN O1** and **ODN O7a** that has not been treated with coupling reagents was purified by the same stepwise method as described in Figure S3B. Lane 1 contains the resulting fraction at 13 minutes (containing the organic modified DNA strands). Lane 2 contains the fraction at 13 minutes according to the HPLC spectrum in Figure S3B. The short dimer is easily observed because all the unmodifed DNA strand are removed and the SYBr gold stained gel can be images by integrating the fluorescent light for 4 seconds.

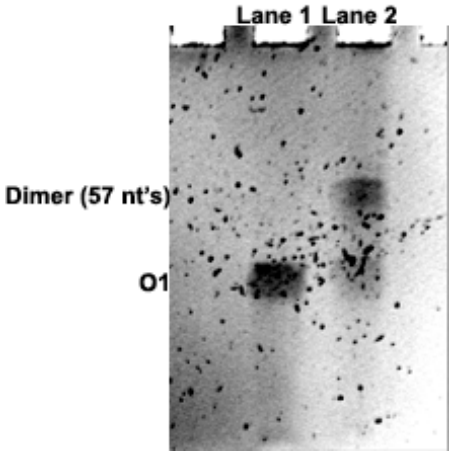


Figure S4. Non-stained 9% denaturing PAGE similar to the gel in Figure 7, but with the dihydrazine as the coupling reagent. The 4-HB incorporating **ODN O1**, **O2**, **O3** and **O4**. Lane 1 (4-HB with **ODN O1**, **O2**, **O3** and **O4** without dihydrazine treatment). Lane 2 (4-HB with **ODNO1** and **O2** treated with dihydrazine). Lane 3 (4-HB with **ODN O1**, **O2** and **O3** treated with dihydrazine). Lane 4 (4-HB with **ODN O2**, **O3** and **O4** treated with dihydrazine). Lane 5 (4-HB with **ODN O1**, **O2**, **O3** and **O4** treated with dihydrazine). Lane 6 (4-HB with **ODN O2** and **O3** treated with dihydrazine). Lane 7 4-HB with **ODN O3** and **O4** treated with dihydrazine). Lane 8 (4-HB with **ODN O1** and **O3** treated with dihydrazine). Lane 9 (4-HB with **ODN O1** and **O4** treated with dihydrazine). The hydrazone dimer consisting of **ODN O1, O2, O3** or **O4** (organic dimer with 84-88 nt) is clearly observed.

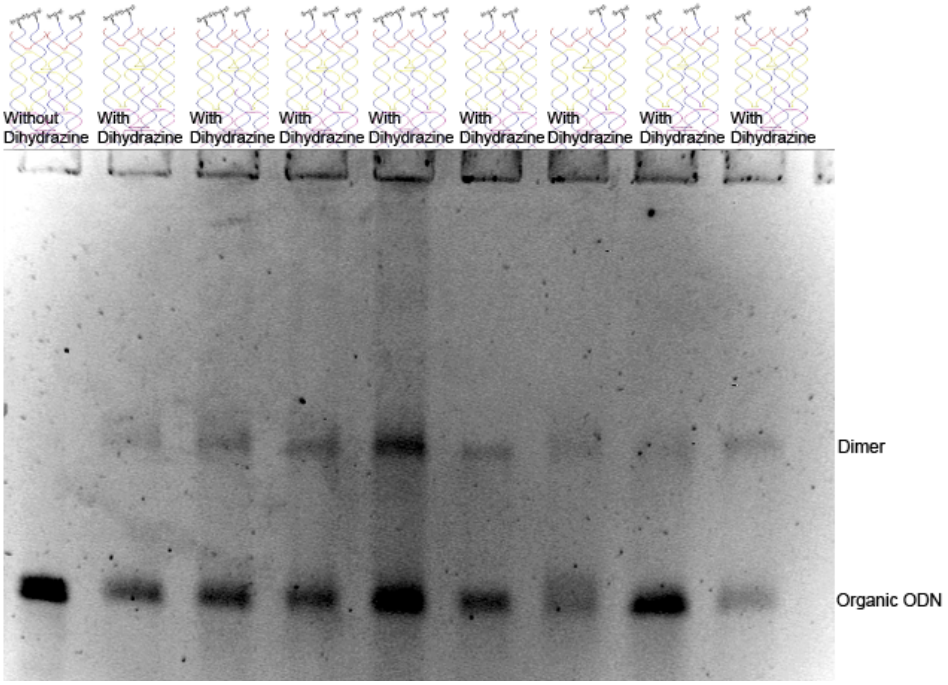
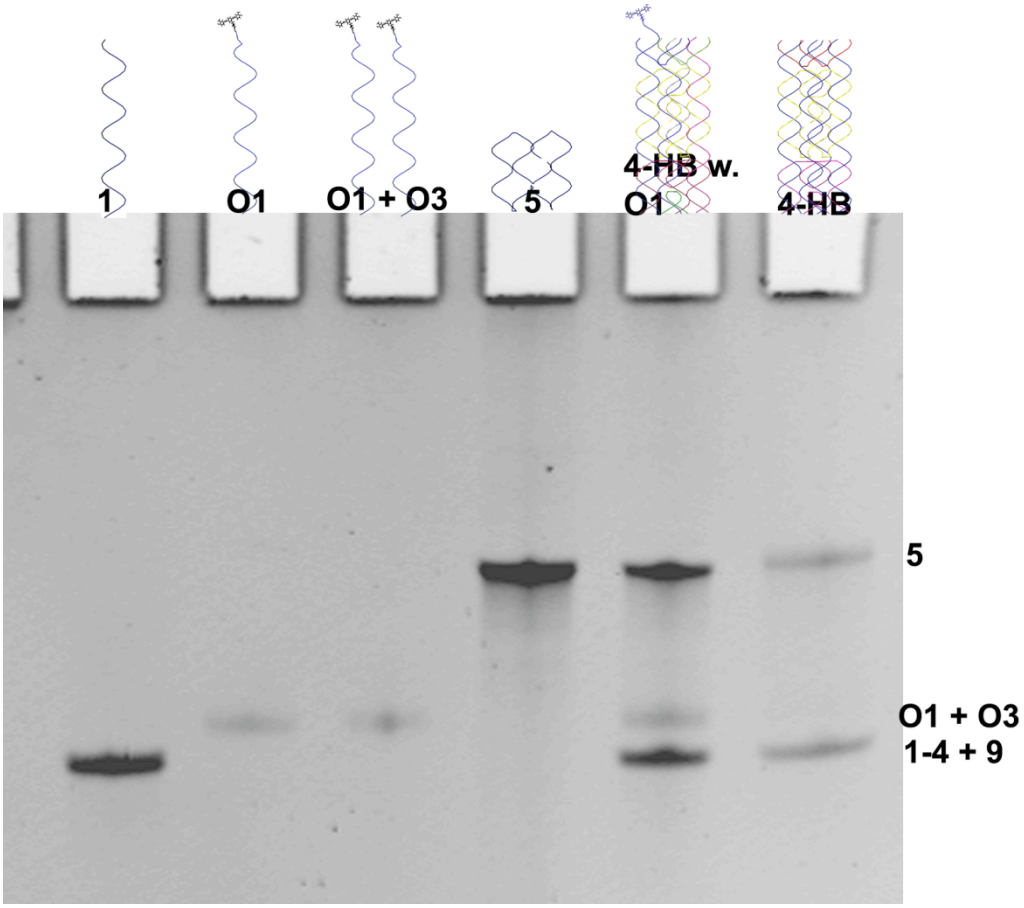
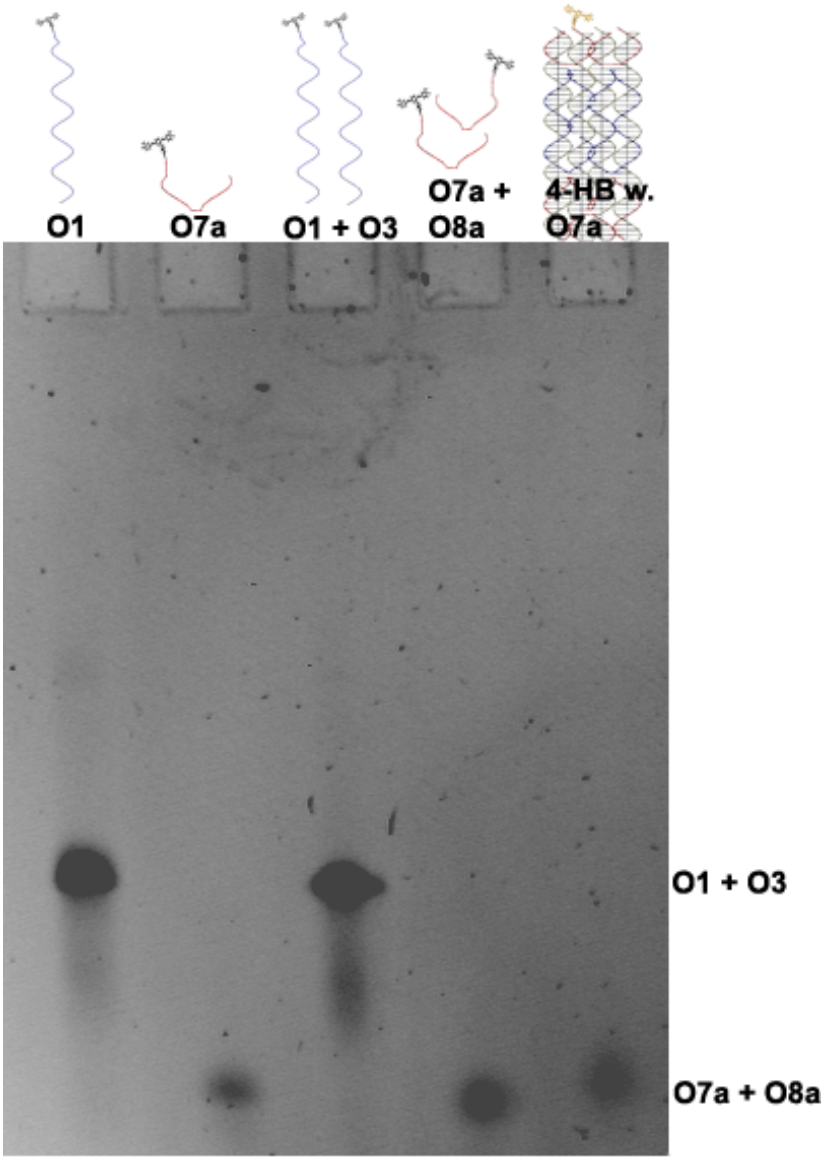


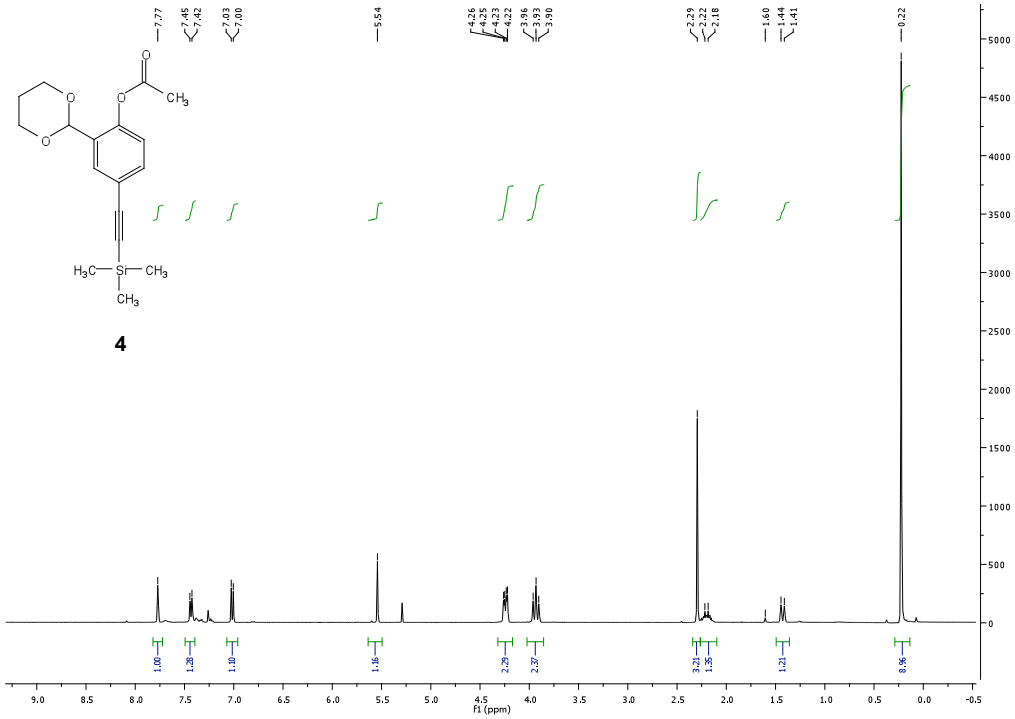
Figure S5. Tests for unspecific couplings in, (A) nickel-salen formation and (B) for dihydrazone formation. (A) Lane 1 (**ODN 1**). Lane 2 (**ODN O1**). Lane 3 (**ODN O1** and **O3**). Lane 4 (**ODN 5**). Lane 5 (4-HB with **O1**). Lane 6 (4-HB). Prior to gel loading was all solutions treated with nickel(II)chloride and EDA. The gel clearly verifies that no intermolecular couplings are observed.



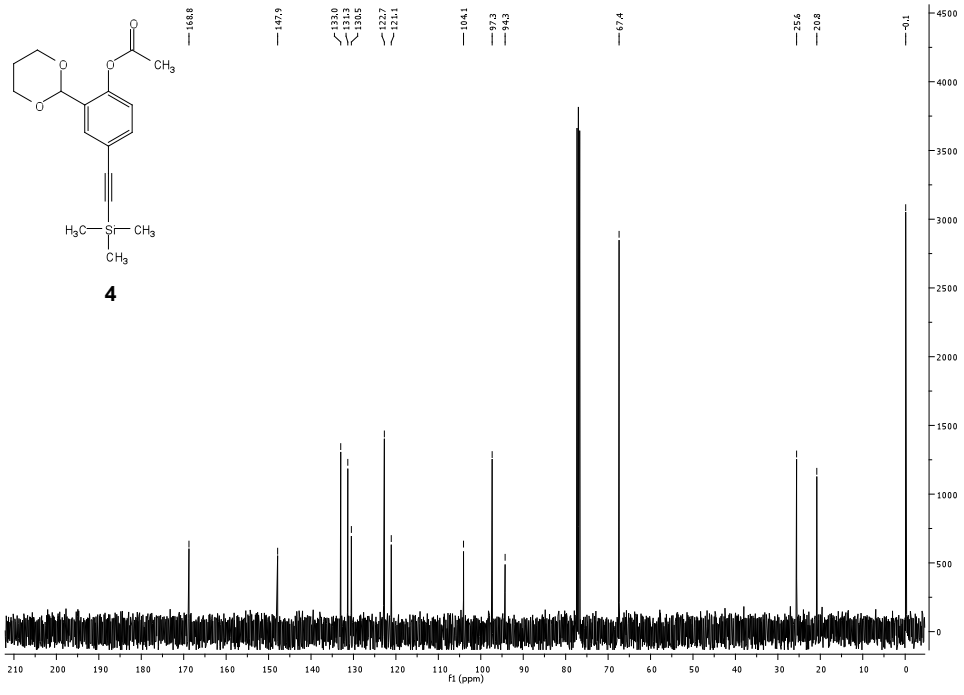
(B) Lane 1 (**ODN 1**). Lane 2 (**ODN O7a**). Lane 3 (**ODN O1** and **O3**). Lane 4 (**ODN O7a** and **O8a**). Lane 5 (4-HB with **O7a**). Prior to gel loading was all solutions treated with dihydrazine for two hours. The gel clearly verifies that no intermolecular couplings are observed.



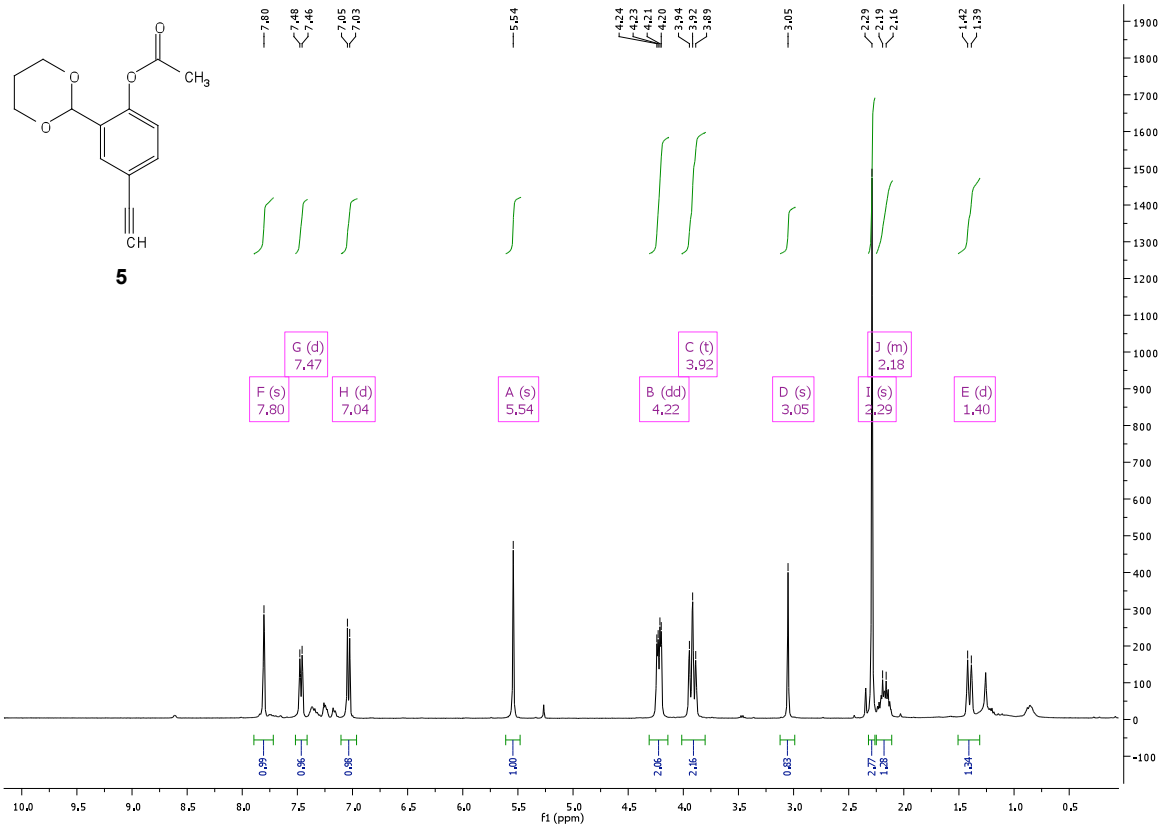
NMR spectra

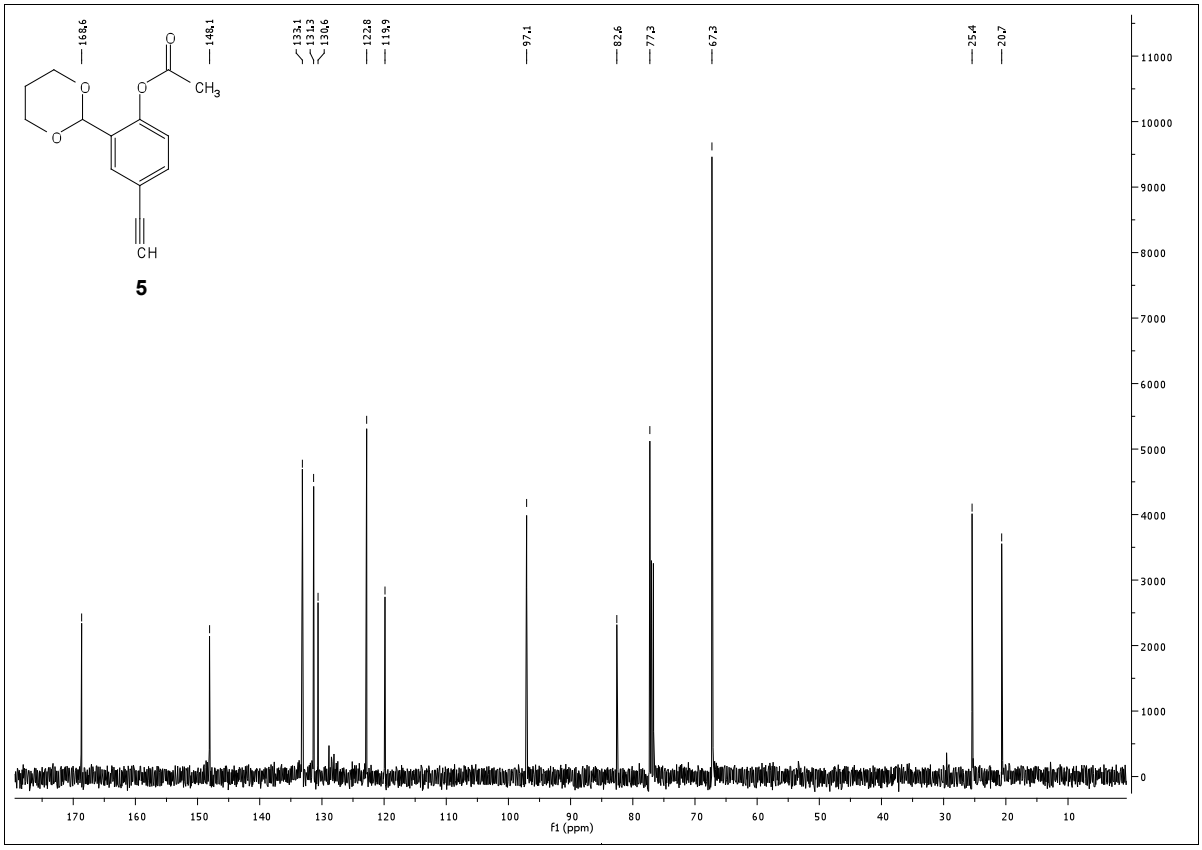


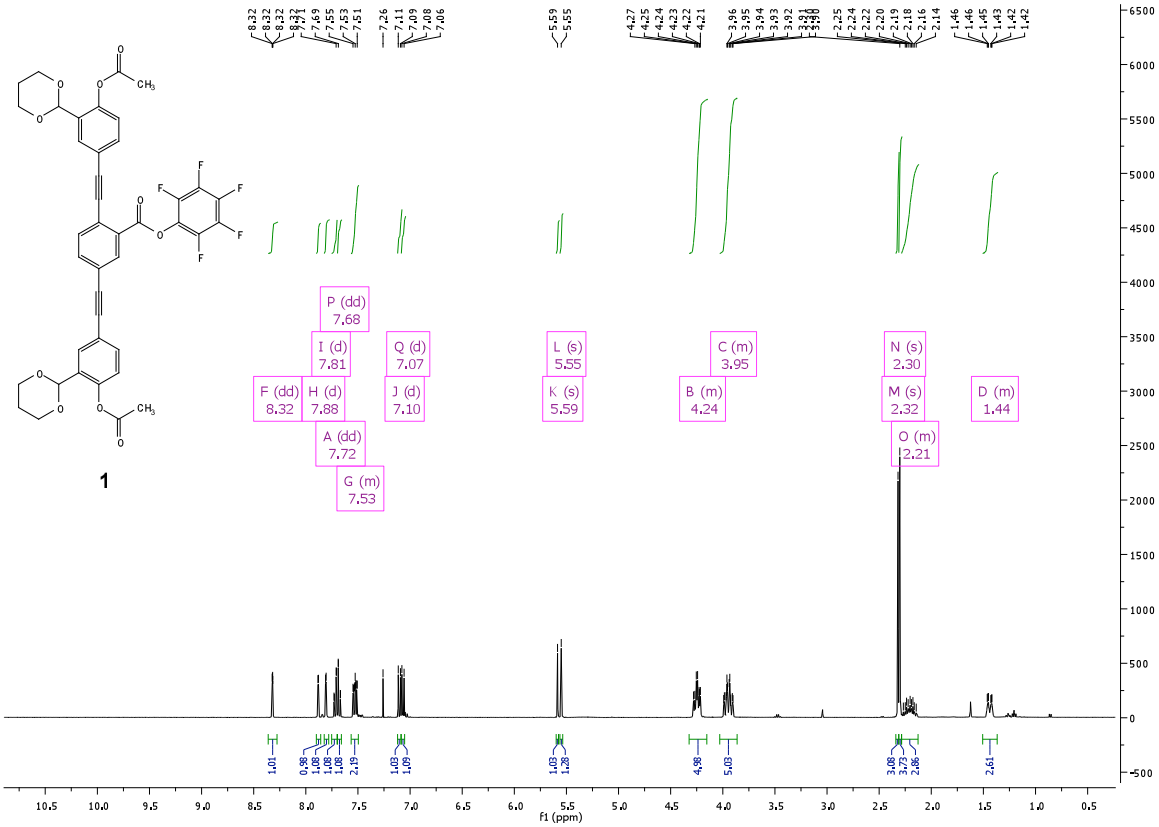
4

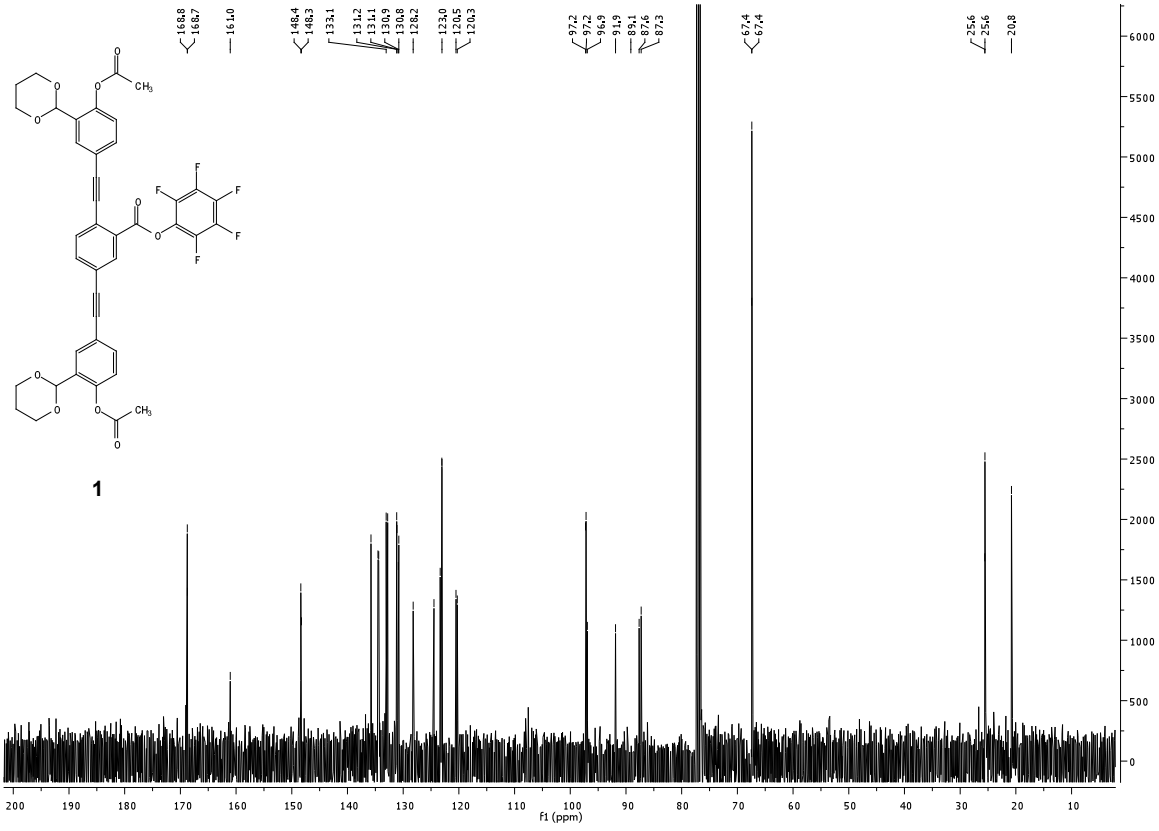


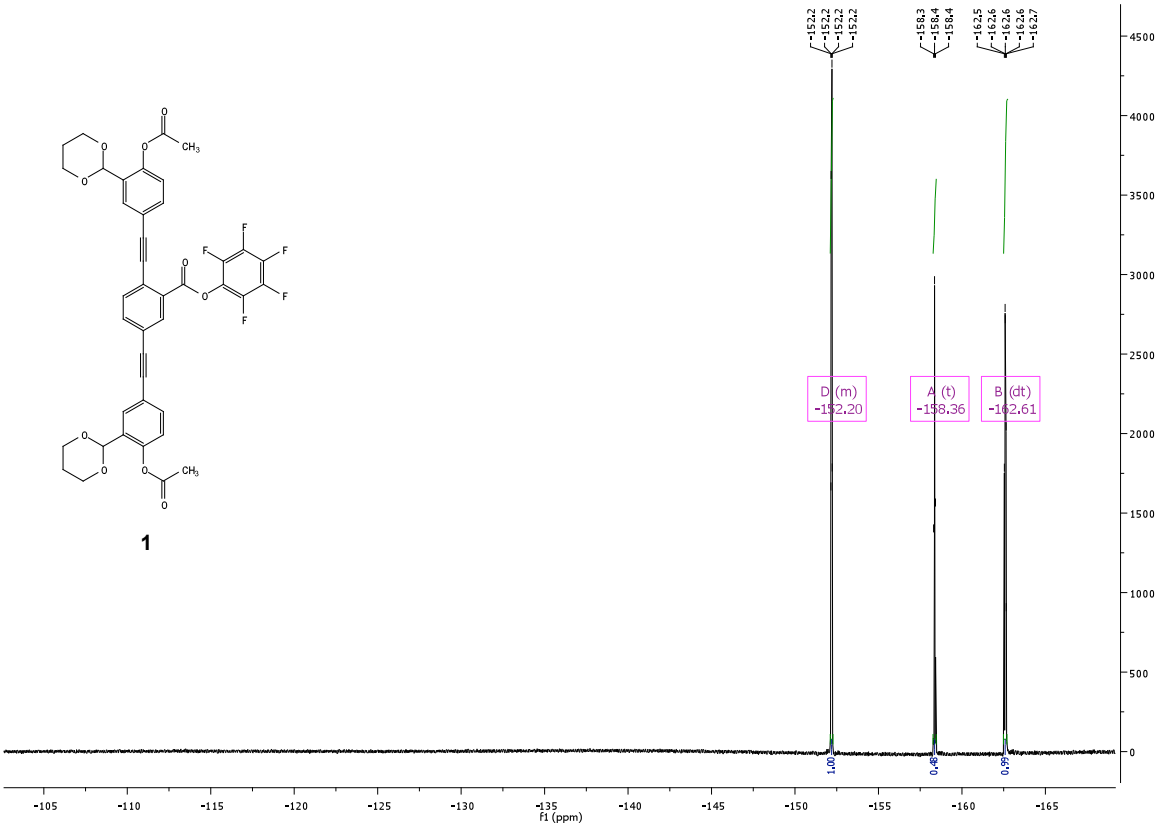
4


5
F (s) 7,80
G (d) 7,47
H (d) 7,04
A (s) 5,54
B (dd) 4,22
C (t) 3,92
D (s) 3,05
I (s) 2,29
J (m) 2,18
E (d) 1,40
f1 (ppm)


5
f1 (ppm)


P (dd) 7.68
I (d) 7.81
Q (d) 7.07
L (s) 5.55
C (m) 3.95
N (s) 2.30
F (dd) 8.32
H (d) 7.88
J (d) 7.10
K (s) 5.59
B (m) 4.24
M (s) 2.32
D (m) 1.44
A (dd) 7.72
O (m) 2.21
G (m) 7.53
1
f1 (ppm)


1
f1 (ppm)


-152.2
-152.2
-152.2
-152.2
-152.2
-158.3
-158.4
-158.4
-158.4
-162.5
-162.6
-162.6
-162.6
-162.7
D (m)
-152.20
A (t)
-158.36
B (dt)
-162.61
1.00
0.48
0.98
4500
4000
3500
3000
2500
2000
1500
1000
500
0
-105
-110
-115
-120
-125
-130
-135
-140
-145
-150
-155
-160
-165
f1 (ppm)

1